Human neurocysticercosis: Comparison of different diagnostic tests in cerebrospinal fluid

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Neurocysticercosis (NC), caused by the larval stage of *Taenia solium*, is one of the most common parasitic diseases of the central nervous system. NC diagnosis is mostly based on costly brain neuroimaging (computed tomography and/or nuclear magnetic resonance) that is rarely accessible in most affected areas. The most sensitive and specific tools for NC diagnosis are imagery techniques. The identification of specific antibodies and antigens are currently used only to support NC diagnosis due to their limited specificity and sensitivity.

This study was performed to compare immunodiagnostic assays (antibody detection by ELISA and EITB and HP10 antigen detection by ELISA) with the detection of parasite DNA by PCR amplification of a repetitive element of the parasite genome in the cerebrospinal fluid (CSF) of 121 radiological and clinically characterized NC patients. Patients were divided into six groups according to the stage of parasites and their localization. CSF cellularity of each patient was also recorded.

When all patients were considered, PCR exhibited the highest sensitivity (95.9%) and variable specificity (80% or 100%) depending on the controls used. Sensitivity of antibody detection by ELISA and EITB was not significantly different and ELISA identified HP10 antigen mostly when vesicular cysticerci were located in the subarachnoidal basal cisterns. These results can help in the selection of different individual assays or their combination to be used in NC diagnosis according to different requirements.

Keywords: Neurocysticercosis, diagnosis, immunological tools, PCR
Neurocysticercosis (NC), caused by the larval stage of the cestode parasite *Taenia solium*, is one of the most common parasitic diseases of the central nervous system (CNS) (20, 39). This disease represents a major public health problem in endemic areas of Latin America, Africa and Asia (18, 29, 33, 36) and has been diagnosed with increasing frequency in the United States, primarily as an imported disease (8). In Mexico, seroprevalence rates for human cysticercosis varied from 3.7 to 12.2% (18) and computerized tomography (CT) based epidemiological studies performed in inhabitants of rural communities have found a prevalence of NC up to 10% (14, 17).

Most NC cases are asymptomatic caused by parasites established in the parenchyma. However, due to the high prevalence of the CNS infection, symptomatic NC is also frequent. Symptomatic NC may adopt different forms, from clinically mild to a severe disabling disease (12). The most frequent clinical NC manifestations are seizures (NC is the most common cause of late-onset epilepsy in endemic countries (28, 35)), intracranial hypertension, neurological deficits and mental changes (3). In these heterogeneous clinical presentations, parasite (location, size, number) and host factors (degree of immune and inflammatory reactions developed) are involved (13, 23). The non-specificity of the neurological symptoms makes it impossible to diagnose the disease on clinical grounds alone. For this reason, in most of cases, NC diagnosis is based on neuroimaging studies (CT, and magnetic resonance imaging, MRI). CT and MRI also provide information with respect to parasite stage and location (21, 31). Three main stages have been described: vesicular (viable), colloidal (degenerative) and calcified (inactive) (19). CT is the best radiological method to detect intra-parenchymal calcification and MRI is more sensitive for the identification of cysts in the ventricles (42). However, in some cases, particularly when parasites are located in the subarachnoid basal cisterns, CT or MRI cannot detect the parasite.
In these cases NC diagnosis is supported by clinical, epidemiological and serological data and the response to the cysticidal treatment (9).

Immunodiagnosis of NC by detecting antigen (Ag) or/and antibodies (Ab) is an accessible low cost diagnostic tool in endemic areas. For Ab detection, the Enzyme-Linked Immunosorbent Assay (ELISA) and the Enzyme-Linked Immunoelectrotransfer Blot (EITB) employing total or partially purified antigen preparations are the most widely used techniques (7, 10). Many authors report that Ab detection in sera identifies approximately 70% of NC patients, the missing cases being those with a single parasite (43) and parenchymal and/or damaged cysticerci (5, 15, 37, 42). It is also estimated that its specificity for detecting active infection is around 30%, mainly due to the persistence of antibodies months or years after resolution of the infection (22). Extra-neural cysticercosis and cross-reaction with other cestodes and helminths can also contribute to false positives detected by using sera antibodies (22, 27). However, better Ab detection is reported when they are detected in the cerebrospinal fluid (CSF) of neurological patients (6). The detection of secreted cysticercal Ag by ELISA is highly sensitive and specific for the diagnosis of living cysticerci (vesicular) localized in the subarachnoidal space at the base of the skull (15, 16). More recently, methods based on the detection of cysticercal DNA have become increasingly explored. In one study, using highly repetitive elements of the parasite as probes, as low as 10 fg of *T. solium* DNA was detected by PCR in the CSF in 29 out of 30 patients (1). In addition, a semi-nested PCR based on HDP2 that can detect 0.174 fg of *T. solium* DNA has also been described (24, 25). Here, a comparative study of immunological procedures plus the procedure described by Almeida et al (1) were evaluated using CSF from neurological patients from Mexico and France.

**Materials and methods**

**Patients and samples**
This study was performed on CSF obtained by lumbar puncture from 121 patients (66 men and 55 women) who were admitted to the Instituto Nacional de Neurología y Neurocirugía (INNN), in Mexico City, Mexico between March 2004 and May 2008. Age at diagnosis ranged from 15 to 72 years (mean 40.3; median 40, interquartile range (IQR) 31-50). The stage (vesicular, colloidal or calcified) and location (parenchyma and basal subarachnoid space or ventricle) of cysticerci was based on CT or/and MRI. CSF cellularity (considered increased when the concentration of white blood cells (WBC) exceeded 5 cells per µl) and hydrocephaly was recorded (clinically defined). A total of 20 CSF samples from non-NC Mexican neurological patients (mainly samples of patients with epilepsy, tumour, demyelinating disease, headache or congenital subarachnoidal cysts) and 49 CSF from non-NC patients (with toxoplasmosis, malaria, HIV or candidosis) in the Parasitology-Mycology Laboratory at the Pitié-Salpêtrière hospital, Paris were included.

Classification of neurocysticercosis cases

All patients included as NC patients were established and confirmed based on radiological features, characteristics of CSF, clinical presentation and evolution and response to treatment. Patients were classified as follows: NC patients with vesicular (group 1), colloidal (group 2) or calcified cysticerci (group 3); patients for whom a doubt existed regarding the presence of a vesicular cyst (group 4); patients included after successful cysticidal treatment for whom, at the moment of sampling, radiological studies did not detect any parasites (group 5). Those patients with vesicular parasites (group 1) were classified according to parasite location: parenchyma or subarachnoid sulci (group 1a) and subarachnoid basal cisterns or ventricles (group 1b). Group 4 corresponds mainly to patients with unilateral enlargement of a basal cistern but without direct evidence of parasites. In this location (subarachnoid basal cisterns), the radiological visualization of the parasite is often difficult to determine since the parasites
exhibited a similar intensity than CSF. The cysticerci appearance usually does not enhance
after the use of gadolinium, and they commonly lack the scolex.

Detection of specific antibodies

Anti-*T. solium* Ab levels were determined by a homebrew ELISA. Vesicular fluid recovered
as previously described (26) from *T. solium* cysticerci was used as the source of Ag. CSF
samples were diluted (1/50) and 100 µl of each sample diluted in PBS-BSA buffer was used.
Samples were run in duplicate and considered positive if the mean of the optical density (OD)
at 450 nm was higher than the cut-off (corresponding to the mean of 5 negative CSF samples
+ 2 SD, ranging from 0.06 to 0.10). Negative samples were from non-NC neurological
patients of INNN diagnosed by MRI (different from our control group). We also included as
positive controls samples from NC patients of the INNN previously diagnosed based on MRI,
lumbar puncture, clinical examination and follow-up.

EITB (LDBIO Diagnostics, Lyon, France) was also performed (40). The procedure
recommended by the manufacturer was used with the following minor modifications for a
better reading of the strips. The detection of at least two bands was indicative of NC. The
membrane strip was incubated 5 min in buffer R2 before adding 50 µl of CSF samples. Strips
were incubated on the rocking platform overnight at room temperature (instead of 90 min).
After washing, strips were incubated an additional 60 min with the anti-IgG conjugate at
room temperature. After washing, strips were incubated with NBT/BCIP substrate in the dark
for 60 min (instead of 10-30 min). The reaction was stopped after aspiration of the liquid by
adding distilled water. We used the positive control provided in the kit and the same negative
controls from the INNN.

Detection of specific antigens
Parasite HP10 Ag was detected by a homebrew ELISA as previously described (16). Samples were performed in duplicate. A sample was considered positive if the mean OD value at 450 nm was greater than the cut-off value (corresponding to the mean of 5 negative CSF samples + 2 SD, ranging from 0.12 to 0.19). The cut-off value was estimate for each plate using five CSF samples from confirmed non-NC neurological patients of INNN diagnosed by MRI (separate from our control group). A group of five additional samples from neurological patients confirmed as NC positive controls from the INNN were included.

Detection of *Taenia solium* DNA

The presence of *T. solium* DNA was explored by PCR in each CSF sample. Primers designed to amplify the highly repetitive element, pTsol9, of the genome were employed (Genbank accession code U45987) (1, 4). This technique can detect 10 fg of DNA as it was previously reported (1).

CSF samples were centrifuged for 10 min at 12,000 rpm. Supernatants were removed and 100 µl of phosphate buffered saline (PBS) were added. Genomic DNA was obtained using a spin column kit (DNeasy blood and tissue kit, Qiagen). *Taenia solium* gDNA obtained from cysticerci of naturally infected pigs using the same procedure was used as a positive control.

Reactions were performed in a final volume of 50 µl, containing 1.5 mM MgCl$_2$, 0.5 µM of each primer, 200 µM of dNTPs (Q-BIOgen), 1.25 U of ampliTaq gold DNA polymerase (Applied Biosystems) and 2 µl of DNA from CSF sample. The primers used in the PCR to amplify pTsol9 were: 5’-CAGGGTGTGACGTCATGG-3’ (forward primer, position 21-38 or 179-196 or 336-353) and 5’-GCTAGGCAACTGGCCTCCT-3’ (reverse primer, position 122-140 or 280-298 or 437-455). As control of amplification, we work also in another tube with 1 µl of DNA from CSF sample plus 1 µl of DNA from pig cysticerci. In the first cycle, DNA was denaturated at 95°C for 5 min, followed by 38 cycles of denaturing at 95°C for 45 sec,
primer annealing at 57°C for 60 sec and elongation at 72°C for 45 s, plus one cycle at 72°C for 10 min. Ten microliters of PCR amplification products were evaluated on 2% agarose gel. A specific amplification product of 120 bp was expected for one repeat unit. When higher amounts of DNA were present, the amplification of 2 and 3 repeat units was also observed (Figure 1). The presence of one band is sufficient to consider the PCR positive. This technique was also evaluated on negative controls from the Pitié-Salpêtrière hospital.

Statistical analysis
Data were processed in Microsoft Excel 2008 and Stata/SE 9.0. The sensitivity and specificity of each tool were determined with the 95% confidence interval. Data were compared using Pearson’s $\chi^2$ test with Yates’s correction when appropriate.

Concordance of tool was estimated with kappa of Cohen with the 95% confidence interval. If $K=1$, the concordance is perfect. Between 0.81 and 1, concordance is excellent; good between 0.61 and 0.8; moderate between 0.41 and 0.6; poor between 0.21 and 0.4; null between 0 and 0.2.

Pearson’s test of correlation was used to evaluate the correlation between the OD values of ELISA tests and the number of cells. The Kruskal-Wallis test was used to determine the difference in mean number of cells compared to results of PCR and EITB. The level of significance was less than 5%.

Ethical considerations
The present study fulfills the regulations for research using human subjects stated by Mexican laws and International regulations. It also complies with all ethical aspects considered in the General Rules of Health for Clinical Investigation. All participants volunteered to enter the
study, donated a sample and gave informed consent. Results were confidential. All patients received medical attention and specific treatment required by a neurologist at the INNN.

Results

Description of patients

Table 1 summarizes the main clinical and biological characteristics of the patients included in this study. Of the 121 confirmed NC patients, 51 had vesicular cysticerci (group 1), 6 colloidal cysticerci (group 2), 31 calcified cysticerci (group 3); in 19 of individuals there was a doubt concerning the presence of vesicular cysticerci (group 4) and 14 did not show cysticerci in the radiological studies at the moment of sampling (group 5, inclusion after adequate response to the cysticidal treatment).

CSF cellularity (Table 1) was recorded in 117 confirmed NC samples and 11 controls. In NC patients, cellularity varied from 0 to 399 cells/mm$^3$ (mean=30.6 $\pm$ 56.4, median=9). Of them 69 (59%) were inflammatory (WBC count > 5/$\mu$l). In controls, the cellularity varied between 0 and 80 (mean=10.9 $\pm$ 26.0, median=2), being inflammatory for 2 of them (22.2%). The number of cells in the CSF was missing in 2 patients with vesicular cysts, 1 with a calcification and 1 without distinguishable cysts on the radiological studies.

Sensitivity and specificity of each diagnostic tool

Table 2 shows the sensitivity and specificity of each technique employed. For diagnosis of vesicular parasites, the sensitivity of the Ag-ELISA was significantly lower than those of the Ab-ELISA and PCR. This difference disappeared when samples were analysed according to parasite location. Sensitivity of Ag-ELISA for diagnosis of colloidal and calcified parasite or when no parasites were not distinguished on the radiological studies but the cysticidal treatment successfully solved the clinical symptoms and the CSF cellularity was also lower.
than in the other techniques. This difference was significant when this test was compared with Ab-ELISA and PCR in cases of calcified parasite or when no parasites were detectable. Compared to the other test, PCR sensitivity was the highest in all groups of patients. Significant differences between PCR and EITB were observed particularly when all patients including those with calcified cysts were considered. Although sensitivity of Ab detection using ELISA was higher than EITB in all patient groups except one (sensitivities were equal in group 4), this difference was not significant (p>0.05). Specificity of the tools varied between 80% for PCR to 100% for Ag-ELISA and EITB with no significant differences between techniques. Of the 20 controls, 4 were positive by PCR (presence of specific band). Of these, one was also positive with the Ab-ELISA. This patient had multiple cavernomas. The PCR was also positive for a sample from a patient, for whom, although having a cyst on brain imaging, the diagnosis of NC was discarded by lumbar puncture result, response to treatment, clinical and radiological evolution assessed by neurological examination and repeated MRI. Another patient who was also positive by PCR had a chronic hydrocephaly but without other clinical signs for NC. When tested on the non-NC French CSF samples, PCR specificity increased to 100 %.

Concordance between diagnostic tools

We evaluated the concordance of each procedure, one by one, in the different groups of patients (Table 3). The value of kappa ranged from 0.11 to 0.89. In Table 3, the best concordance found was between the Ab-ELISA and the EITB test. Between these techniques, the concordance was excellent for groups 1, 1b and 4, good for groups 2, 3 and 5 and moderate only for the vesicular parenchymal cysticerci group (group 1a). The poorest concordance was between Ag-ELISA and PCR, being in 4 of the 7 patient categories poor or null as shown in Table 3. It must be noted the low concordance of Ag-ELISA compared to the
other techniques regarding groups 3 and 5. In these cases, only 1 concordance was moderate, the other 5 were poor or null. The best concordance between each tool was observed for group 1b.

CSF cellularity and sensitivity of diagnostic tests

The number of cells (mainly lymphocytes) in the CSF positively correlated with the level of antibody \( r=0.42, p<0.001 \) and the level of HP10 Ag \( r=0.24, p=0.006 \). Also, PCR and EITB positive samples showed significantly higher numbers of cells than negative ones \( p=0.0007 \) and \( p=0.0001 \), respectively. None of the CSF samples were hemorrhagic.

Discussion

PCR based on pTsol9 amplification detected from 90 to 100% of NC cases depending on the stage and location of the parasite. Unexpectedly, this technique detected 100% of parenchymal NC. This observation is not in agreement with previous data that indicate that parenchymal cysticerci have a restricted access to the CSF (15). The specificity of the PCR using Mexican controls was 80% and 100% using French samples. This latter result is in accordance with those reported by Almeida et al (1). The specificity obtained with Mexican samples was not expected as this procedure is based on specific amplification of parasite DNA. One sample among the four CSF samples that were PCR positive was also positive for the detection of antibodies by ELISA. It is very probable that these positive controls had or have had a NC that was not diagnosed by radiological techniques. As we stated above, undetected infections could occur in endemic conditions without apparent symptoms or radiological signs. PCR could be useful to diagnose the NC cases when imagery techniques have failed.
In the study on HDP2 PCR, two uncertain NC cases were PCR positive (25). The authors suggest that parasite DNA detected in these cases could be related to the perilesional oedema occasionally observed with damaged cysticerci (30). Indeed, we have no information concerning the process leading to DNA release in the CSF. Further studies are required to better understand this process and also to evaluate the persistence of DNA in the CSF of patients.

The results obtained for the other immunological tools are in agreement with previous reports. A specificity of 100% was observed with the HP10 Ag-ELISA, accompanied by a high sensitivity to detect vesicular parasites located in the subarachnoid space or in ventricles (86.4%). We also confirmed that its capacity to detect vesicular parenchymal cysts is limited, although higher than that reported previously (42.9% vs 0%) (2). These contradictory results could be influenced by the low number of parenchymal NC included in both studies. In addition, a radiological misdiagnosis of undistinguishable additional subarachnoid cysts cannot be discarded. Particularly, in our study, 2 of the 3 positive patients showed inflammatory CSF, a feature mostly seen when parasites were localized in subarachnoid basal space.

In general, positive samples using different diagnostic tools were significantly more inflammatory than negative ones showing the relevance of the presence of parasite markers in the genesis of the immune-inflammatory reaction associated. This was found in previous studies in which inflammatory CSF was found to be associated with the positive level of HP10 Ag with Odds Ratios of 30 and 32 respectively (2, 16).

Results reported for the Ab-ELISA regarding detection of vesicular parasites (sensitivity = 97.7% when located in the subarachnoid space or ventricles and 85.7% when located in the parenchyma) were similar to previous studies (15). It must also be noted that this technique effectively detects parasites independently of their viability.
The sensitivity and specificity of the EITB were initially described as 98 and 100% respectively (41). A subsequent evaluation of this test in 50 NC patients found a sensitivity of 91% for active NC and 88% for calcified cysticerci (43). Our results were not significantly different from this latter report (90.2% and 61.3% respectively). In our study, although EITB exhibited a higher specificity than Ab-ELISA (100% vs. 90%, respectively) and a lower sensitivity (81% vs. 90.1%, respectively), these differences were not statistically significant. Our result differs from some published studies (34, 38) although others are in agreement (11, 32). Since our Ab-ELISA is a simpler and less expensive tool for the immunological diagnosis of NC in the CSF, it seems more advisable for use in poor endemic countries to consolidate the diagnosis of NC.

PCR in CSF samples remains to be evaluated for the post-treatment follow-up. It will be also interesting to evaluate PCR in serum samples because CSF sampling needs a lumbar puncture. Serum samples are more readily available but contain a lot of proteins and DNAs that can limit the specificity of the PCR.

In summary, comparing the procedures currently in use to NC diagnosis encourage the use of parasite DNA detection by PCR for diagnosis and point to the existence of possible undiagnosed NC cases not detectable by radiological or immunological available tests.

Acknowledgements

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Table 1: Principal characteristics of patients and controls included in the study

<table>
<thead>
<tr>
<th></th>
<th>N</th>
<th>Male/Female</th>
<th>Age (median)</th>
<th>Number of cells/mm³ (median)</th>
<th>Hydrocephaly</th>
<th>Ab-ELISA</th>
<th>Ag-ELISA</th>
<th>EITB</th>
<th>PCR</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Confirmed NC</strong></td>
<td>121</td>
<td>66/55</td>
<td>41 (19-72)</td>
<td>9 (0-399)</td>
<td>44</td>
<td>109</td>
<td>12</td>
<td>65</td>
<td>98</td>
</tr>
<tr>
<td><strong>Group 1</strong></td>
<td>51</td>
<td>27/24</td>
<td>40 (19-72)</td>
<td>19 (0-399)</td>
<td>26</td>
<td>49</td>
<td>2</td>
<td>41</td>
<td>10</td>
</tr>
<tr>
<td><strong>Group 1a</strong></td>
<td>7</td>
<td>4/3</td>
<td>38 (22-56)</td>
<td>4 (0-53)</td>
<td>0</td>
<td>6</td>
<td>1</td>
<td>3</td>
<td>4</td>
</tr>
<tr>
<td><strong>Group 1b</strong></td>
<td>44</td>
<td>23/21</td>
<td>42 (19-72)</td>
<td>25.5 (0-399)</td>
<td>26</td>
<td>43</td>
<td>1</td>
<td>38</td>
<td>6</td>
</tr>
<tr>
<td><strong>Group 2</strong></td>
<td>6</td>
<td>3/3</td>
<td>44.5 (28-51)</td>
<td>6.5 (0-12)</td>
<td>0</td>
<td>6</td>
<td>0</td>
<td>2</td>
<td>4</td>
</tr>
<tr>
<td><strong>Group 3</strong></td>
<td>31</td>
<td>11/20</td>
<td>39 (21-67)</td>
<td>3 (0-85)</td>
<td>5</td>
<td>23</td>
<td>8</td>
<td>7</td>
<td>24</td>
</tr>
<tr>
<td><strong>Group 4</strong></td>
<td>19</td>
<td>14/5</td>
<td>46 (28-69)</td>
<td>24 (0-180)</td>
<td>9</td>
<td>18</td>
<td>1</td>
<td>12</td>
<td>18</td>
</tr>
<tr>
<td><strong>Group 5</strong></td>
<td>14</td>
<td>11/3</td>
<td>41 (23-58)</td>
<td>4 (0-176)</td>
<td>4</td>
<td>13</td>
<td>1</td>
<td>3</td>
<td>11</td>
</tr>
<tr>
<td><strong>Control (Mexico)</strong></td>
<td>20</td>
<td>9/11</td>
<td>28 (15-58)</td>
<td>2 (0-80)</td>
<td>ND</td>
<td>2</td>
<td>18</td>
<td>0</td>
<td>20</td>
</tr>
<tr>
<td><strong>Control (Paris)</strong></td>
<td>49</td>
<td>32/17</td>
<td>49 (17-74)</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>49</td>
</tr>
</tbody>
</table>

Group 1=vesicular, 1a=vesicular in parenchyma or subarachnoid sulci, 1b=vesicular in subarachnoid basal cisterns or ventricles, 2=colloidal, 3=calcified, 4=patients with doubts regarding the presence of a vesicular cyst, 5=patients included after successful cysticidal treatment; Ab-ELISA= antibody detection by ELISA, Ag-ELISA= antigen detection by ELISA, POS=positive, NEG=negative, ND= not done
Table 2: Sensitivity and specificity of the tested diagnostic tools in the different patient categories.

<table>
<thead>
<tr>
<th>Patient Category</th>
<th>Sensitivity</th>
<th>Specificity</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>NAb-ELISA</td>
<td>NAg-ELISA</td>
</tr>
<tr>
<td>Confirmed NC</td>
<td>121</td>
<td>90.1 [83.3-94.8]</td>
</tr>
<tr>
<td>Group 1</td>
<td>51</td>
<td>96.1 [86.5-99.5]</td>
</tr>
<tr>
<td>Group 1a</td>
<td>7</td>
<td>85.7 [42.1-99.6]</td>
</tr>
<tr>
<td>Group 1b</td>
<td>44</td>
<td>97.7 [88-99.9]</td>
</tr>
<tr>
<td>Group 2</td>
<td>6</td>
<td>100 [54.1-100]</td>
</tr>
<tr>
<td>Group 3</td>
<td>31</td>
<td>74.2 [55.4-88.1]</td>
</tr>
<tr>
<td>Group 4</td>
<td>19</td>
<td>94.7 [74.9-99.9]</td>
</tr>
<tr>
<td>Group 5</td>
<td>14</td>
<td>92.9 [66.1-99.9]</td>
</tr>
</tbody>
</table>

P1: Ab-ELISA vs. Ag-ELISA, P2: Ab-ELISA vs. EITB, P3: Ab-ELISA vs. PCR, P4: Ag-ELISA vs. EITB, P5: Ag-ELISA vs. PCR, P6: EITB vs. PCR.

Group 1=vesicular, 1a=vesicular in parenchyma or subarachnoid sulci, 1b=vesicular in subarachnoid basal cisterns or ventricles, 2=colloidal, 3=calcified, 4=patients with doubts regarding the presence of a vesicular cyst, 5=patients included after successful cysticidal treatment.
Table 3: Concordance of tools estimated by kappa of Cohen with 95% confidence intervals

<table>
<thead>
<tr>
<th></th>
<th>Ab-ELISA vs Ag-ELISA</th>
<th>Ab-ELISA vs EITB</th>
<th>Ab-ELISA vs PCR</th>
<th>Ag-ELISA vs EITB</th>
<th>Ag-ELISA vs PCR</th>
<th>EITB vs PCR</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Confirmed NC</strong></td>
<td>0.38 *</td>
<td>0.70 *</td>
<td>0.50 *</td>
<td>0.49 *</td>
<td>0.21 *</td>
<td>0.41 *</td>
</tr>
<tr>
<td></td>
<td>(0.26-0.49)</td>
<td>(0.64-0.88)</td>
<td>(0.32-0.68)</td>
<td>(0.36-0.62)</td>
<td>(0.10-0.31)</td>
<td>(0.25-0.58)</td>
</tr>
<tr>
<td><strong>Group 1</strong></td>
<td>0.70 *</td>
<td>0.84 *</td>
<td>0.70 *</td>
<td>0.70 *</td>
<td>0.54 *</td>
<td>0.67 *</td>
</tr>
<tr>
<td></td>
<td>(0.53-0.86)</td>
<td>(0.70-0.97)</td>
<td>(0.57-0.92)</td>
<td>(0.65-0.94)</td>
<td>(0.36-0.73)</td>
<td>(0.48-0.85)</td>
</tr>
<tr>
<td><strong>Group 1a</strong></td>
<td>0.46 *</td>
<td>0.58 *</td>
<td>0.60 *</td>
<td>0.51 *</td>
<td>0.31 *</td>
<td>0.40 *</td>
</tr>
<tr>
<td></td>
<td>(0.10-0.89)</td>
<td>(0.24-0.93)</td>
<td>(0.29-0.91)</td>
<td>(0.29-0.39)</td>
<td>(0.20-0.60)</td>
<td>(0.1-0.71)</td>
</tr>
<tr>
<td><strong>Group 1b</strong></td>
<td>0.76 *</td>
<td>0.89 *</td>
<td>0.77 *</td>
<td>0.87 **</td>
<td>0.62 *</td>
<td>0.74 *</td>
</tr>
<tr>
<td></td>
<td>(0.60-0.92)</td>
<td>(0.77-1)</td>
<td>(0.59-0.94)</td>
<td>(0.74-0.99)</td>
<td>(0.43-0.82)</td>
<td>(0.57-0.92)</td>
</tr>
<tr>
<td><strong>Group 2</strong></td>
<td>0.31 *</td>
<td>0.68 *</td>
<td>0.68 *</td>
<td>0.19 *</td>
<td>0.25 *</td>
<td>0.55 *</td>
</tr>
<tr>
<td></td>
<td>(0.07-0.62)</td>
<td>(0.31-0.71)</td>
<td>(0.36-0.94)</td>
<td>(0.34-0.49)</td>
<td>(0.02-0.26)</td>
<td>(0.24-0.87)</td>
</tr>
<tr>
<td><strong>Group 3</strong></td>
<td>0.28 *</td>
<td>0.76 *</td>
<td>0.42 *</td>
<td>0.42 *</td>
<td>0.11 *</td>
<td>0.37 *</td>
</tr>
<tr>
<td></td>
<td>(0.10-0.76)</td>
<td>(0.59-0.94)</td>
<td>(0.18-0.65)</td>
<td>(0.19-0.66)</td>
<td>(0.02-0.25)</td>
<td>(0.16-0.59)</td>
</tr>
<tr>
<td><strong>Group 4</strong></td>
<td>0.59 *</td>
<td>0.69 *</td>
<td>0.69 *</td>
<td>0.68 *</td>
<td>0.11 *</td>
<td>0.49 *</td>
</tr>
<tr>
<td></td>
<td>(0.36-0.93)</td>
<td>(0.76-1)</td>
<td>(0.47-0.92)</td>
<td>(0.46-0.90)</td>
<td>(0.20-0.73)</td>
<td>(0.48-0.91)</td>
</tr>
<tr>
<td><strong>Group 5</strong></td>
<td>0.22 *</td>
<td>0.69 *</td>
<td>0.71 *</td>
<td>0.38 *</td>
<td>0.16 *</td>
<td>0.54 *</td>
</tr>
<tr>
<td></td>
<td>(0.04-0.44)</td>
<td>(0.45-0.93)</td>
<td>(0.48-0.94)</td>
<td>(0.06-0.69)</td>
<td>(0.02-0.33)</td>
<td>(0.29-0.79)</td>
</tr>
</tbody>
</table>

*a excellent concordance (0.81-1), b good (0.61-0.8), c moderated (0.41-0.6), d poor (0.21-0.4), e null (0-0.2)

Group 1=vesicular, 1a=vesicular in parenchyma or subarachnoid sulci, 1b=vesicular in subarachnoid basal cisterns or ventricles, 2=colloidal, 3=calcified, 4=patients with doubts regarding the presence of a vesicular cyst, 5=patients included after successful cysticidal treatment
Figure 1: Amplicons from PCR after separation by electrophoresis in agarose gel.

The lane contained molecular weight markers (M), samples of cerebrospinal fluid from patients (lane 1-16) and negative control (T). For each sample, two amplifications are realized: the first one with 2 µl of DNA from CSF sample and the second one with 1 µl of DNA from CSF sample plus 1 µl of DNA from pig cysticerci. In the second one, we observed 2 repeat units. All samples were positives except sample 10.
Human Neurocysticercosis: Comparison of Different Diagnostic Tests Using Cerebrospinal Fluid

Lorraine Michelet, Agnès Fleury, Edda Sciutto, Eric Kendjo, Gladis Fragoso, Luc Paris, and Bernard Bouteille

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